Citation:

Allen RR, Carson L, Kwik-Uribe C, Evans EM, Erdman JW Jr. Daily consumption of a dark chocolate containing flavanols and added sterol esters affects cardiovascular risk factors in a normotensive population with elevated cholesterol. *J Nutr.* 2008 Apr;138(4):725-31.

PubMed ID: 18356327

Study Design:

Randomized Crossover Trial

Class:

A - Click here for explanation of classification scheme.

Research Design and Implementation Rating:

POSITIVE: See Research Design and Implementation Criteria Checklist below.

Research Purpose:

To examine the effect of the regular consumption of a flavanol-containing chocolate bar with added phytosterols on serum cholesterol levels in a free-living population and to assess the effect of the dietary treatments on blood pressure and selected markers of inflammation and adhesion molecules.

Inclusion Criteria:

- Male or female aged 24-70 years
- Fasting serum total cholesterol levels between 5.20 - 7.28 mmol/L
- Body Mass Index (BMI) of 20 - 40 kg/m^2

Exclusion Criteria:

- Taking cholesterol-lowering, anti-hypertensive, weight loss, or other drugs which affect blood lipids within 6 months of the study
- Use of herbal supplements, including antioxidant and sterol-containing supplements
- Hypertension defined as systolic > 159 mm Hg and diastolic > 99 mm Hg
- History of other chronic diseases including type 1 or 2 diabetes mellitus and cardiovascular disease
- Pregnant or planning to become pregnant

Description of Study Protocol:

Recruitment

Subjects were recruited from flyers, posters, email, electronic messages and mail through the University of Illinois campus and local area physicians, hospitals and clinics.

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Design

Double-blind, placebo-controlled randomized crossover study with all subjects following a lead in 2 week diet and then 4 weeks each of intervention and control.

Blinding used

Double-blind trial.

Intervention

Subjects consumed 2 chocolate bars per day as part of the American Heart Association "An Eating Plan for Healthy Americans" encouraged diet.

Chocolate bars were 22 grams cocoa flavanol containing dark chocolate with (1.1 g canola sterol esters) and without plant sterols.

Statistical Analysis (SPSS version 14.0)

- Means, standard deviation and distribution statistics
- Student's t test
- Repeated measures ANOVA
- Power analysis

Data Collection Summary:

Timing of Measurements

- Baseline measurements taken after 2 week lead in diet phase
- 4 weeks of either treatment or control
- Final at end of second 4 week period crossover treatment or control

Dependent Variables

- Serum lipids including: total cholesterol, low density lipoprotein (LDL), very low density lipoprotein (VLDL), high density lipoprotein (HDL), and triacylglycerol, mmol/L
- High sensitivity c-reactive protein (hs-CRP), mg/L
- Glycosylated hemoglobin (HbA1c), %
- Soluble CD40 ligand (sCD-40L), µg/L
- Intercellular adhesion molecule (ICAM-1), µg/L
- Blood pressure, mm Hg

Independent Variables

- Subjects consumed 2 chocolate bars per day as part of the American Heart Association "An Eating Plan for Healthy Americans" encouraged diet.
- Chocolate bars were 22 grams cocoa flavanol containing dark chocolate with (1.1 g canola sterol esters) and without plant sterols.

Control Variables

Description of Actual Data Sample:

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Initial N: 650 interested subjects from recruiting measures, 49 entered study

Attrition (final N): 44 subjects completed study (PS+ group, 66% male; PS- group, 64% male)

Age: PS+, 45.9 ± 8.1 years; PS-, 43.5 ± 8.9 years

Ethnicity: not described

Other relevant demographics: not described

Anthropometrics: BMI PS+, 28.1 $\pm$ 4.6 kg/m^2; PS-, 27.4 $\pm$ 4.6 kg/m^2

Location: Champagne Urbana area of Illinois, USA

Summary of Results:

Key Findings

- Total cholesterol reduced 3% in the Plant Sterol (PS+) group compared to the non-plant sterol (PS-) group (p=0.017).
- LDL cholesterol reduced 4% in the PS+ compared to PS- (p=0.014).
- Cocoa flavanols (CF) reduced systolic and diastolic blood pressures regardless of plant sterol content (systolic 8.2%, p<0.001 at 4 week and 5%, p<0.001 at 8 week and diastolic 8.2%, p<0.001 at week 4, 3.4%,p<0.05 at 6 week and 2.2%, p=0.09 at 8 week).

Author Conclusion:

The regular consumption of flavanol-containing chocolate bar with added plant sterols as part of a low-fat diet can significantly lower flood cholesterol levels and systolic blood pressure.

Reviewer Comments:

Strength: double blind crossover design with detailed data collection

Supported by grant from Mars, Inc. Hacketts town, NJ.

Research Design and Implementation Criteria Checklist: Primary Research

Relevance Questions

1.	Would implementing the studied intervention or procedure (if found successful) result in improved outcomes for the patients/clients/population group? (Not Applicable for some epidemiological studies)	Yes
2.	Did the authors study an outcome (dependent variable) or topic that the patients/clients/population group would care about?	Yes
3.	Is the focus of the intervention or procedure (independent variable) or topic of study a common issue of concern to nutrition or dietetics practice?	Yes

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4.	Is the intervention or procedure feasible? (NA for some epidemiological studies)	Yes

Validity Questions

1.	**Was the research question clearly stated?**	Yes
1.1.	Was (were) the specific intervention(s) or procedure(s) [independent variable(s)] identified?	Yes
1.2.	Was (were) the outcome(s) [dependent variable(s)] clearly indicated?	Yes
1.3.	Were the target population and setting specified?	Yes
2.	**Was the selection of study subjects/patients free from bias?**	Yes
2.1.	Were inclusion/exclusion criteria specified (e.g., risk, point in disease progression, diagnostic or prognosis criteria), and with sufficient detail and without omitting criteria critical to the study?	Yes
2.2.	Were criteria applied equally to all study groups?	Yes
2.3.	Were health, demographics, and other characteristics of subjects described?	Yes
2.4.	Were the subjects/patients a representative sample of the relevant population?	Yes
3.	**Were study groups comparable?**	Yes
3.1.	Was the method of assigning subjects/patients to groups described and unbiased? (Method of randomization identified if RCT)	Yes
3.2.	Were distribution of disease status, prognostic factors, and other factors (e.g., demographics) similar across study groups at baseline?	Yes
3.3.	Were concurrent controls used? (Concurrent preferred over historical controls.)	Yes
3.4.	If cohort study or cross-sectional study, were groups comparable on important confounding factors and/or were preexisting differences accounted for by using appropriate adjustments in statistical analysis?	N/A
3.5.	If case control or cross-sectional study, were potential confounding factors comparable for cases and controls? (If case series or trial with subjects serving as own control, this criterion is not applicable. Criterion may not be applicable in some cross-sectional studies.)	N/A
3.6.	If diagnostic test, was there an independent blind comparison with an appropriate reference standard (e.g., "gold standard")?	N/A
4.	**Was method of handling withdrawals described?**	Yes

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4.1.	Were follow-up methods described and the same for all groups?	Yes
4.2.	Was the number, characteristics of withdrawals (i.e., dropouts, lost to follow up, attrition rate) and/or response rate (cross-sectional studies) described for each group? (Follow up goal for a strong study is 80%.)	Yes
4.3.	Were all enrolled subjects/patients (in the original sample) accounted for?	Yes
4.4.	Were reasons for withdrawals similar across groups?	Yes
4.5.	If diagnostic test, was decision to perform reference test not dependent on results of test under study?	N/A
5.	**Was blinding used to prevent introduction of bias?**	Yes
5.1.	In intervention study, were subjects, clinicians/practitioners, and investigators blinded to treatment group, as appropriate?	Yes
5.2.	Were data collectors blinded for outcomes assessment? (If outcome is measured using an objective test, such as a lab value, this criterion is assumed to be met.)	Yes
5.3.	In cohort study or cross-sectional study, were measurements of outcomes and risk factors blinded?	N/A
5.4.	In case control study, was case definition explicit and case ascertainment not influenced by exposure status?	N/A
5.5.	In diagnostic study, were test results blinded to patient history and other test results?	N/A
6.	**Were intervention/therapeutic regimens/exposure factor or procedure and any comparison(s) described in detail? Were interveningfactors described?**	Yes
6.1.	In RCT or other intervention trial, were protocols described for all regimens studied?	Yes
6.2.	In observational study, were interventions, study settings, and clinicians/provider described?	N/A
6.3.	Was the intensity and duration of the intervention or exposure factor sufficient to produce a meaningful effect?	Yes
6.4.	Was the amount of exposure and, if relevant, subject/patient compliance measured?	Yes
6.5.	Were co-interventions (e.g., ancillary treatments, other therapies) described?	Yes
6.6.	Were extra or unplanned treatments described?	N/A
6.7.	Was the information for 6.4, 6.5, and 6.6 assessed the same way for all groups?	Yes
6.8.	In diagnostic study, were details of test administration and replication sufficient?	N/A

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7.	**Were outcomes clearly defined and the measurements valid and reliable?**	Yes
7.1.	Were primary and secondary endpoints described and relevant to the question?	Yes
7.2.	Were nutrition measures appropriate to question and outcomes of concern?	Yes
7.3.	Was the period of follow-up long enough for important outcome(s) to occur?	Yes
7.4.	Were the observations and measurements based on standard, valid, and reliable data collection instruments/tests/procedures?	Yes
7.5.	Was the measurement of effect at an appropriate level of precision?	Yes
7.6.	Were other factors accounted for (measured) that could affect outcomes?	Yes
7.7.	Were the measurements conducted consistently across groups?	Yes
8.	**Was the statistical analysis appropriate for the study design and type of outcome indicators?**	Yes
8.1.	Were statistical analyses adequately described and the results reported appropriately?	Yes
8.2.	Were correct statistical tests used and assumptions of test not violated?	Yes
8.3.	Were statistics reported with levels of significance and/or confidence intervals?	Yes
8.4.	Was "intent to treat" analysis of outcomes done (and as appropriate, was there an analysis of outcomes for those maximally exposed or a dose-response analysis)?	No
8.5.	Were adequate adjustments made for effects of confounding factors that might have affected the outcomes (e.g., multivariate analyses)?	Yes
8.6.	Was clinical significance as well as statistical significance reported?	Yes
8.7.	If negative findings, was a power calculation reported to address type 2 error?	Yes
9.	**Are conclusions supported by results with biases and limitations taken into consideration?**	Yes
9.1.	Is there a discussion of findings?	Yes
9.2.	Are biases and study limitations identified and discussed?	Yes
10.	**Is bias due to study's funding or sponsorship unlikely?**	No
10.1.	Were sources of funding and investigators' affiliations described?	Yes
10.2.	Was the study free from apparent conflict of interest?	No

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